# FOULANT ANALYSIS IN CROSS-FLOW FILTRATION USING RESISTANCE-IN-SERIES MODEL

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A dissertation submitted in partial fulfillment of the
requirements for the award of the degree of
Bachelor of Chemical Engineering (Biotechnology)

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APRIL 2010

# ABSTRACT

Membrane technologies grow rapidly in the world and were applied in many industrial processes as an efficient separation technique and for the purpose of wastewater treatment. Membrane separation process was used to replace the conventional method to reduce time of operation, reduce space requirement for downstream process and increase the productivity. There are several types of membrane separation used in the industries which are ultrafiltration, microfiltration, nanofiltration and reverse osmosis. These membrane separations used different types of membrane such as flat sheet, hollow fiber and other. Even though membrane separation has many advantages, it also has a problem which is membrane fouling that resulting in reduces permeate flux and increase the production cost. This study is undertaken to identify and determine the resistances that lead membrane fouling by using Resistance-In-Series Model and Darcy's Law. A cross-flow filtration with polysulfone hollow fiber membrane of molecular pore size 0.2µm was used to separate the buffer solution containing *Escherichia coli*. From the data obtained, the weak adsorption resistance was explored as the main factor resulting membrane fouling in this study. For the membrane recovery, the alkaline cleaning can recover the fouled membrane about 99.8% and it could help increasing the permeate flux for separation process. *E. coli* K-12 strain was identified which can give high permeate flux compared to *E. coli* B strain. Thus, *E. coli* K-12 can be retained by membrane in the separation of succinic acid from fermentation broth.

# ABSTRAK

Teknologi membran membangun dengan pesat di dunia dan telah digunakan dalam proses industri sebagai teknik pemisahan yang berkesan dan digunakan untuk tujuan merawat air sisa. Proses pemisahan membran digunakan untuk menggantikan kaedah konvensional bagi mengurangkan masa operasi, mengurangkan keperluan ruang untuk proses akhir dalam penghasilan dan meningkatkan produktiviti. Terdapat beberapa jenis membran yang digunakan dalam industri seperti penapis ultra, penapis mikro, penapis nano dan osmosis songsang. Terdapat pelbagai jenis membran yang digunakan dalam proses pemisahan seperti jenis kepingan, serat berlubang dan lain-lain. Walaupun proses pemisahan membran memberi banyak kelebihan namun ia juga mempunyai masalah kegagalan membran yang mengakibatkan penghasilan fluks yang rendah dan meningkatkan kos pengeluaran. Kajian ini dilakukan untuk mengenal pasti dan menentukan halangan yang menyebabkan kegagalan membran berlaku dengan menggunakan Model *Resistance-In-Series* dan Hukum *Darcy*. Penapisan cara arah bertentangan dengan menggunakan membran serat berongga polisulfon bersaiz 0.2µm untuk memisahkan larutan yang mengandungi *Escherichia coli*. Daripada data yang diperolehi daripada proses analisis, halangan jerapan lemah dikenalpasti sebagai faktor utama yang menyebabkan kegagalan membran dalam kajian ini. Untuk memulihkan membran, pembersihan menggunakan larutan alkali dapat mengembalikan membran yang tercemar sebanyak 99.8% dan dapat membantu meningkatkan fluks untuk proses pemisahan. *E. coli* K-12 dikenalpasti yang dapat memberikan fluks tinggi berbanding *E. coli* B. Oleh itu, *E. coli* K-12 dapat ditahan oleh membran dalam pemisahan asid suksinik daripada pati proses penapaian.

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| | | |
|---|---|---|
| DI | - | deionized water |
| *E. coli* | - | *Escherichia coli species* |
| EDTA | - | ethylenediaminetetraacetic acid |
| TF | - | tangential flow |
| h | - | hour |
| $HNO_3$ | - | nitric acid |
| kg | - | kilogram |
| kHz | - | kilohertz |
| kPa | - | kilopascal |
| LB | - | Luria Bertani |
| M | - | molar |
| NaOH | - | sodium hydroxide |
| P | - | pressure |
| PEP | - | phosphoenolpyruvate |
| PES | - | polyethersulfone |
| pH | - | potential for hydrogen ion concentration |
| PPHF | - | polypropylene hollow fiber |
| rpm | - | revolution per minute |
| TMP/$\Delta$P | - | transmembrane pressure |
| < | - | less than |

*roman, Greek, superscript, subscript*

# CHAPTER 1

# INTRODUCTION

## 1.1 Background of Study

Succinic acid also known as amber acid or butanedioic acid. It is a dicarboxylic acid with the molecular formula of $C_4H_6O_4$. The first purification of succinic acid from amber by Georgius Agricola is in the year of 1546. After that, it has been produced by microbial fermentation for the use in agricultural, food and pharmaceutical industries (Song and Lee, 2006). Succinic acid is a dicarboxylic acid of potential industrial interest as a platform chemical and very valuable intermediate value chemical with a moderate market. Most of commercial available succinic acid is produced by chemical process which use liquefied petroleum gas (LPG) or petroleum oil as a starting material. As a commodity chemical, succinic acid could replace many commodities based on benzene and intermediate petrochemicals, resulting in a large reduction in pollution from the manufacturer and in the consumption of over 250 benzene-derived chemicals (Zeikus *et al*., 1999). Figure 1.1 illustrates a potential map of the routes leading to succinic-acid-based intermediate and specialty chemicals.

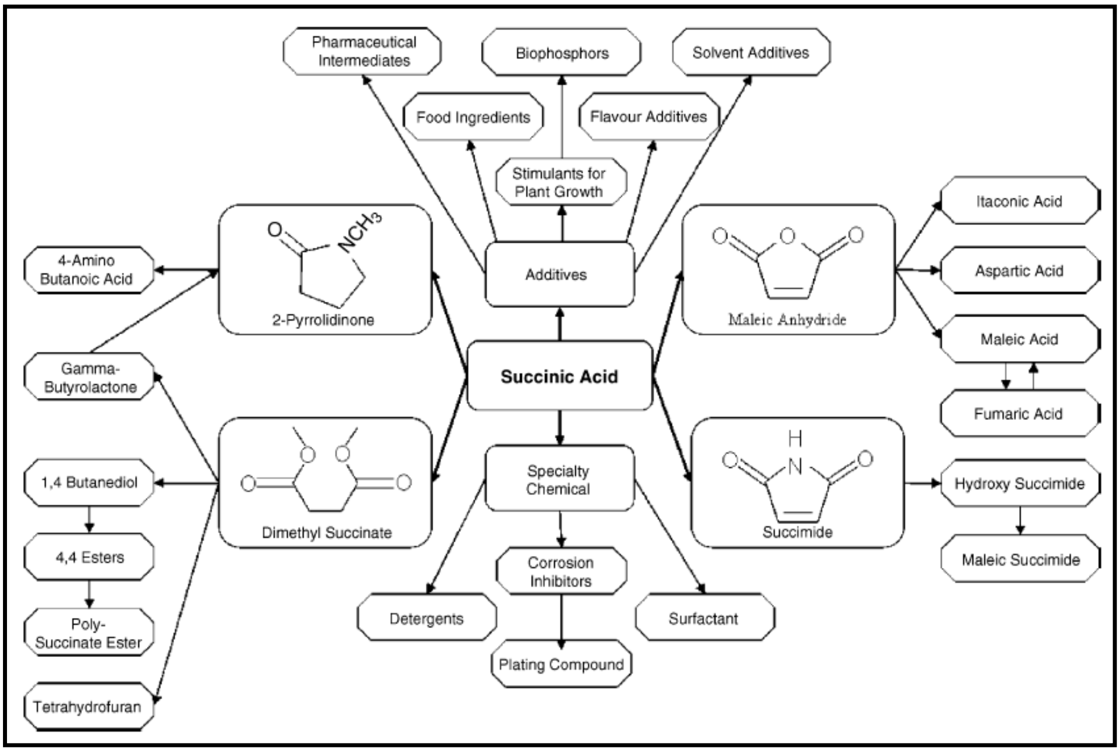


**Figure 1.1** Various chemicals and products can be produced from succinic acid.

Many different microorganisms have been screened and studied for succinic acid production from various carbon sources. *Escherichia coli sp.* is currently used as efficient succinic acid production by being modified in metabolic engineering (Song and Lee, 2006). It is known that *E. coli* under anaerobic conditions produces a mixture of organic acids. In order to obtain a cost-effective production it is necessary to metabolically engineer the organism to produce succinic acid in greater yield than the other acids. The wild type of *E. coli* B strain is used for the production of succinic acid. For increase the efficiency, the mutant type of *E. coli* K-12 strain is produced by researchers in metabolic engineering. Comparison of the K-12 and B strain shows about 3793 proteins of their basic strains are identical although 310 be functional in either K-12 or B but not in both (Studier *et al*., 2009). Formation of by-products in the fermentation process such as acetic, formic and lactic acids is a major problem that has to be solved because it reduces the succinic acid yield and productivity, while increases the complexity and cost of succinic acid recovery.

For the recovery and recycling of primary resources, membrane processes can offer good opportunities (Das and De, 2009). Separation and purification of succinic acid by membrane technology have been studied by researcher to replace current precipitation methods and their associated waste disposal problems. Membrane techniques are widely used for separation purposes; many methods of these processes exist (Kozlowski and Sliwa, 2008). For the fermentation process in the production of succinic acid, the membrane bioreactor could be used to ease separation of microorganism used from liquid product. It is very important to recycle the usage of microorganism in the fermentation because it can reduce the operation cost Thus, the filtration process will be done in small space and can save the downstream time.

Filtration is one of pressure-driven membrane techniques for the separation of dissolved and suspended matter according to their sizes and molecular scales which has been widely, applied in various chemical and biochemical processes. The characteristics of filtration process that makes it excellent for practical applications include the minimized physical damage of biomolecules from shear effects, minimal denaturation, high recovery, high throughput, and cost effectiveness, and the avoidance of resolubilization problems because the solutes can be retained in the solution phase. Compared to the dead-end mode applied mainly in laboratory tests, cross-flow mode has been actually used in continuous operations for the separation of bioproducts such as peptides, proteases, proteins and antibiotics (Juang *et al*., 2008).

While handling with the membrane separation, there are some of disadvantages of using it. The main disadvantage of membrane separation is reduction of permeate flux with time due to membrane fouling. Membrane fouling is caused by the accumulation of feed components on the membrane surface due to concentration polarization and adsorption inside the pores causing pore blocking (Das and De, 2009). An effective method to prevent membrane fouling has not yet been recognized. In the cross-flow filtration with a high feed velocity, the pore blocking and gel layers on the membrane surface are removed by shearing action and a high permeate flux can be maintained. The membrane cleaning and backwashing step are capable to restore the purity of membrane. There are some method to maintain membrane flux performance such as sonication, chemical cleaning and shearing action.

In this research paper, the main focus is about foulant analysis in the cross-flow filtration. The resistances or foulant such as pore blocking and gel formation that cause membrane fouling will be analysed using Resistance-In-Series model. The membrane cleaning also been studied by using chemical cleaning and hydraulic cleaning. The effective cleaning method is identified to restore the membrane flux performance to enhance the separation process. Then, this research paper ends with future prospects for the study of membrane performance in separation process.

## 1.2 Problem Statement

Membrane separation technologies widely used in the industrial application for the downstream process of organic acid production and wastewater treatment. These new technologies with membrane can reduce the space requirement and easiest for the maintenance. There are variety type of membrane separation used in practises which are ultrafiltration, microfiltration, reverse osmosis, gas separation and others. Some advantages can be mentioned for the membrane processes which give the quality of the purified permeate, the moderate operating temperatures and low energy requirements in general, the absence of chemicals, and the fact that it can be combined with other separation processes to enhance the product. Because of these advantages, the chemical industries now are focusing on the usage of membrane to replace the conventional method. The membrane technologies also can reduce the time requirement for the downstream process.

Despite of the many application, membrane technology cannot wide spread use because of the problem such as membrane fouling. The main disadvantage is the flux decline through the membrane during the process, caused by the concentration polarization and the fouling of the membrane. This fouling can be due to the formation of a solid cake onto the membrane surface, the deposition of macromolecules within the membrane pores which are blocked by the solute, and the adsorption of the solutes onto the walls of the membrane (Acero *et al.*, 2009). Membrane fouling wills effects the separation process by reduction of permeate flux through the membrane, as a result of increased flow resistance due to pore blocking, concentration polarization, and cake formation (Lim and Bai, 2003). Reduction of membrane lifetime because of long term effect lead to irreversible fouling from microbial action on the membrane and regeneration of membrane will increase the cost production at downstream process.

Many researchers are searching the method to eliminate membrane fouling and other problem such as using the combination of polymer while fabricating the membrane module and study the factors that affects the membrane fouling. Besides, the membrane cleaning was introduced in the industrial application for membrane separation in order to restore the membrane permeability that effects from pore blockage and other. The chemical cleaning was widely used because of the efficient and significance in the membrane recovery process.

## 1.3 Objective

The main objective of this research is to study the various resistances that lead membrane fouling in cross-flow filtration using Resistances-In-Series model.

## 1.4 Scope of Study

In order to achieve the objective, the following scopes have been identified:

a) The hollow fiber membrane with a molecular pore size of 0.2 µm and area of 110 $cm^2$ was used.
b) Different type of *Escherichia coli* strain used which are B-strain and K12 strain.
c) The phosphate buffer solution for 2 L feed tank will be prepared at optimum pH of 6.5.
d) Foulant analysis to determine various resistances are calculated using Darcy's Law.

## 1.5 Significant of Study

The membrane technologies currently boom up as a new technology that can be used for the separation and purification in the production of organic acid. Besides, the membrane also can be used in the wastewater treatment that can give better treatment compared to other conventional methods such as precipitation and adsorption. As a new technologies, there are many research in the area of membrane was done by the researchers to enhance and grow up the benefit from this technologies. The significant of the research are focusing on development of membrane using combined polymer, the flow of feed patent and to minimize the effect of membrane fouling that can increase the production cost.

For this study, the focus will be on the reduction of membrane fouling to increase the permeate flux, increase the life span of membrane and increase the productivity from effective separation process by studying using *E. coli* strain and resistance in series that were the main factors leads to membrane fouling. The membrane cleaning for the membrane recovery also will be discussed.

CHAPTER 2

# LITERATURE REVIEW

## 2.1 Succinic Acid

Succinic acid, a dicarboxylic acid having the molecular formula of $C_4H_6O_4$, has been used as a precursor for many industrial. Succinic acid is an important chemical as shown in Figure 2.1 (Lee *et al*., 2008). Succinic acid and its derivatives are widely used in foods, pharmaceuticals, and cosmetics. Nowadays, there is increasing demand for its use as a new biodegradable polymer such as polysuccinate (Hong *et al*., 2000).

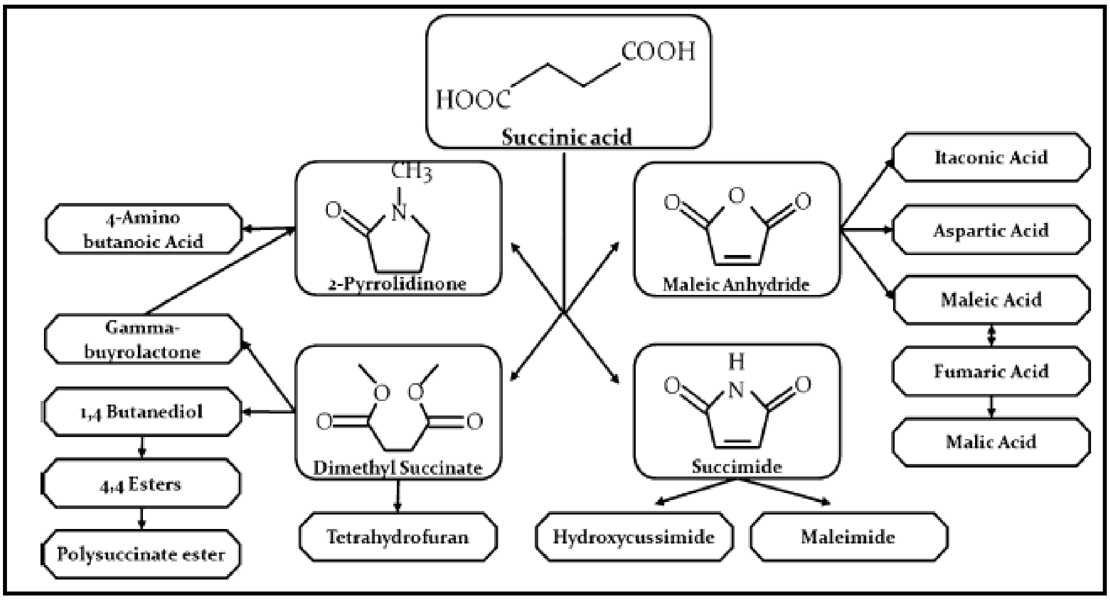


**Figure 2.1** Succinic acid and its derivatives.

Succinic acid is mainly obtained from the hydration of succinic anhydride synthesized from maleic anhydride. Recently, production by fermentation has drawn interest as an alternative (Hong *et al*., 2000). Other potential uses of succinic acid include succinylation of lysine residues to improve the physical and functional attributes of soy proteins in foods, producing modified succinimides used as fuel constituents and in water purification, in the succinylation of cellulose to improve water absorbitivity and the use of succinylated starch as a thickening agent (Zeikus *et al*., 1999).

The production of succinic acid and most of its derivatives is currently at the advanced research and development stage. Succinic acid is also an intermediate for green chemicals and materials. For example, diethyl succinate is a useful solvent for cleaning metal surfaces or for paint stripping, and ethylene- diaminedisuccinate is a replacement for EDTA. Because of its structure as a linear saturated dicarboxylic acid, succinic acid can be used as an intermediate chemical and be converted to 1,4-butanediol, tetrahydrofuran, c-butyrolactone, and other four carbon chemicals that have a world-wide market in excess of $275x10^6$ kg/year. Major advances have been made in the chemical catalysis technology needed to convert succinic acid into these intermediate chemicals. Currently, more than 15,000 tons of succinic acid is manufactured yearly and is sold at US$5.90–8.80/kg, depending on its purity (Zeikus *et al*., 1999).

## 2.2 Succinic Acid Producer

### 2.2.1 *Escherichia coli sp.*

Previous research has shown that *E. coli* B and K-12 are fairly closely related to one another, relative to the tremendous diversity that exists within this species as a whole (Schneider *et al*., 2002). Under anaerobic conditions *E. coli* is known to produce a mixture of organic acids and ethanol. *E. coli* has two hydrophobic membranes surrounding its cytoplasm, the cytoplasmic (or inner) membrane and the outer membrane.(Andersson, 2007) Also, there has been much effort in developing recombinant *Escherichia coli* strains which are capable of enhanced succinic acid production under aerobic and anaerobic conditions (Song and Lee, 2006). The outer membrane contains special proteins, porins, which allow glucose and other small solutes to diffuse through the membrane into the periplasm (the volume between the inner and outer membrane). Figure 2.2 shows the *E. coli* on Luria Bertani agar.

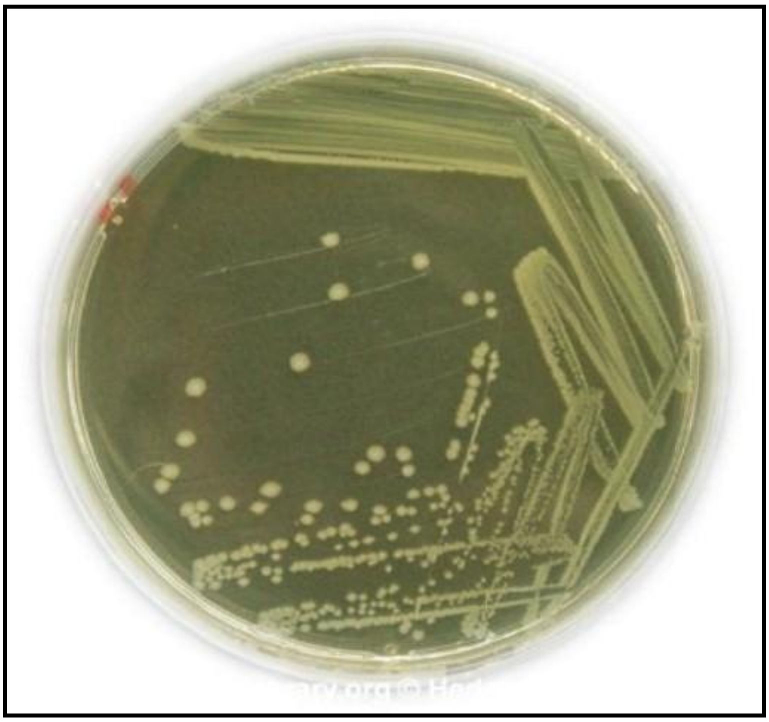

**Figure 2.2** *Escherichia coli sp*. on Luria Bertani agar.

A wild type *E. coli* primarily ferments glucose to ethanol, formic, acetic and lactic acids with only detectable amounts of succinic acid under anaerobic condition. The succinic acid yield on glucose typically obtainable is no more than 0.2 mol $mol^{-1}$. It has been known that *E. coli* utilizes six pathways to form succinic acid, and differently from three bacteria mentioned above, the PEP carboxykinase plays a minor role (Song and Lee, 2006).

The first metabolic engineering approach taken in *E. coli* for efficient succinic acid production was the over expression of the PEP carboxykinase (*pck*) and PEP carboxylase (*ppc*) genes (Song and Lee, 2006). Over expression of the PEP carboxylase resulted in a 3.5 times increase in the amount of succinic acid, whereas the PEP carboxykinase over expression had no effect.

Recently, more effort has been made to develop recombinant *E. coli* strains capable of producing succinic acid with high efficiency. The *Sorghum vulgare ppc* and *Lactococcus lactis pyc* genes were introduced into the *ldh–pfl*-inactivated and *ldh–pta–ack*-inactivated *E. coli* mutant strains respectively, in order to redirect the accumulated pyruvate to oxaloacetic acid (Song and Lee, 2006).

Although metabolically engineered strains of *E. coli* were remarkable in their performance compared with the wild type strain, it's still need improvement. In particular, the specific and volumetric succinic acid productivities are much lower than those obtained with *A. succiniciproducens* and rumen bacteria (Song and Lee, 2006).

### 2.2.2 *Anaerobiospirillum succiniciproducens*

The most investigated organism is *A. succiniciproducens*, a strict anaerobe. *A.succiniciproducens* has been characterised in a number of studies both with regards to medium composition and processing conditions (Andersson, 2007). One of the achievements with *A. succiniciproducens* was conversion of wood hydrolysates into succinate with a mass yield of 0.88 gram succinate per gram glucose. The main drawback with the organism is that it does not seem to tolerate succinic acid concentrations higher than 30-35 g $L^{-1}$.

### 2.2.3 *Mannheimia succiniciproducens*

Recently a facultative anaerobe, *M. succiniciproducens*, was isolated from bovine rumen and shown to produce succinate as its main fermentation product at yields in the order of 0.7 gram succinate per gram sugar consumed. This organism has been demonstrated to ferment both glucose and xylose from wood hydrolysates. The highest succinic acid concentration obtained in *M. succiniciproducens* fermentations is 52.4 g $L^{-1}$. The concentration was achieved using LB growth medium. *M. succiniciproducens* seems promising, and future work should focus on development of mutants able to produce succinate in higher yields (Andersson, 2007).

### 2.2.4 *Actinobacillus succinogenes*

*A. succinogenes* is also a facultative anaerobe inhabiting the bovine rumen (Andersson, 2007). The organism has been shown to produce succinic acid to impressive concentrations (105.8 g $L^{-1}$) in good yields (approximately 0.85 gram per gram glucose). The fermentations that have reached succinate concentrations of more than 80 g $L^{-1}$ have been neutralised with $MgCO_3$. When using $NH_4OH$ or sodium alkali final concentrations were in the range of 60 g $L^{-1}$, which is lower than the concentration some *E. coli* strains have been shown to produce. Unlike *E. coli* or *A. succiniciproducens, A. succinogenes* is a moderate osmophile and has good tolerance to a high concentration of glucose, which is beneficial for fermentation (Song and Lee, 2006).

## 2.3 Mode of Filtration

### 2.3.1 Cross-flow Filtration

Among several filtration methods, dead-end and cross-flow filtrations are the most frequently used in industry. Pressure-driven membrane processes such as ultrafiltration and microfiltration have been widely applied in various chemical and biochemical processes for the separation of dissolved and suspended matter according to the size and molecular scale (Juang *et al*., 2008). Usually, most of the separation of microorganisms from fermentation broth is performed by centrifugation. In cross-flow filtration, the main stream of the suspension is along a direction parallel to the filter medium. The filtrate flow is perpendicular to the main stream and the cake formed is of a limited thickness (Mota *et al*., 2002). Recently, cross-flow microfiltration has been used to separate cells in continuous fermentation processes. The long term performance of membrane units at high cell densities is affected by the fouling of filtration membranes, which require extensive cleaning protocols (Li *et al*., 2006).

In cross-flow filtration, the direction of feed flow is tangential to the membrane surface. As a result, accumulation of filtered solids can be minimized by the sharing action of the flow (Li *et al*., 2006). Cross-flow membrane filtration has been used as an alternative for the separation of soluble intracellular protein from cell lysates (Bailey and Meagher, 2000). Inclusion bodies have also been separated from the soluble proteins in *E. coli* cell lysates using cross-flow microfiltration. A successful lactic acid recovery approach has been that of continuous fermentation in a cell-recycled reactor where the cells are separated by a filtration unit and returned to the fermenter while the product is removed in the permeate (Li *et al*., 2006). Figure 2.3 shows the schematic process of cross-flow filtration system.

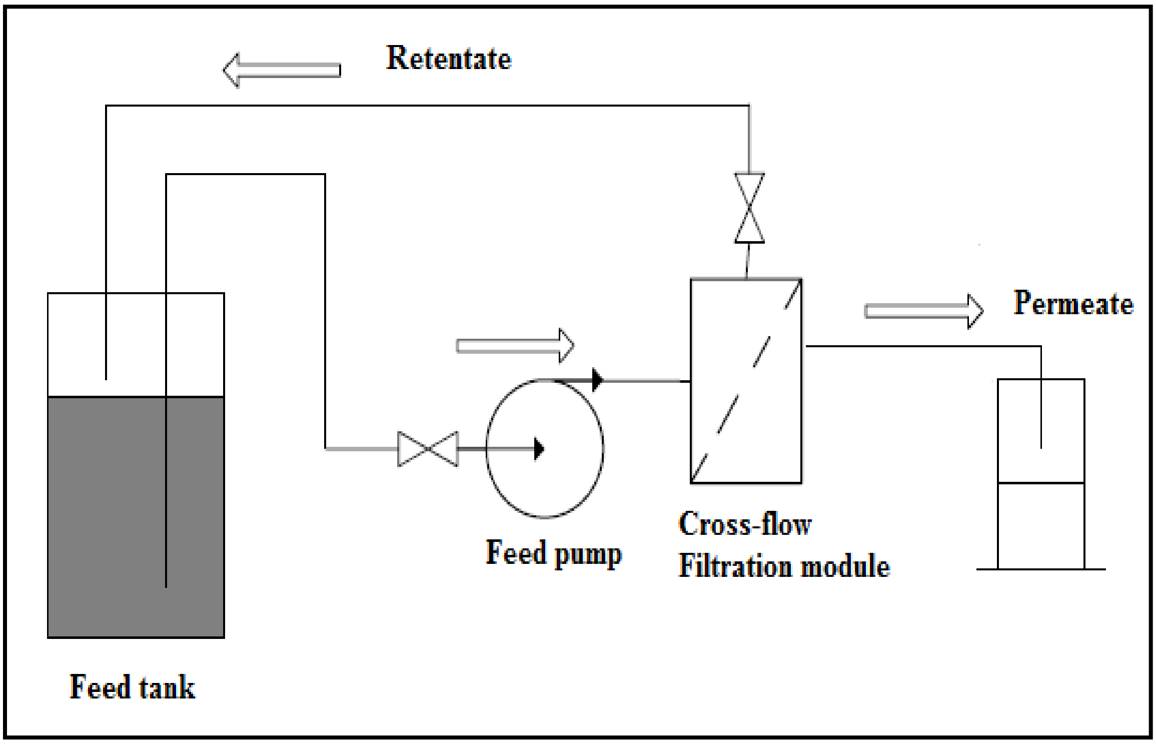


**Figure 2.3** Schematic diagram of the cross-flow filtration system.

Most studies on the use of cross-flow membrane filtration have focused on permeate flux and protein transmission with little regard to the importance of membrane cleaning. Ease and efficiency of membrane cleaning are important in terms of developing a lower cost process (Bailey and Meagher, 2000). This is particularly true for low cost commodity products such as industrial enzymes, antibiotics, alcohols and organic acids. Typical cleaning agents following processing of cell lysates are detergents with and without enzymes, sodium hydroxide, acids and, for resistant membranes, sodium hypochlorite. Elevated temperatures are also usually recommended (Bailey and Meagher, 2000).

### 2.3.2 Dead End Filtration

The dead-end filtration consists in gradually increasing the cake thickness up to a level determined by pressure drop or flow velocity. In the end, a cake and a clarified filtrate will be obtained. Dead-end filtration is often used as a method to estimate the specific cake resistance for cross-flow filtration and usually gives reasonable data for spherical and ellipsoidal-shaped cells. The cake resistance of the layer formed by rod-like particles (bacteria), contrary to the yeast cell layer, displays significant differences in dead end and cross-flow filtration (Mota *et al*., 2002).

A number of studies have been carried out with a view of constructing mathematical models appropriate to description of the filtration behavior of dead-end hollow fiber membranes for which cake resistance increases during filtration (Chang *et al*., 2006). The change in the filtration resistance caused by cake formation on the membrane surface was estimated by assuming a linear relationship between the specific resistance and pressure drop across the cake layer. Effects of the expanded double layer around particle surfaces on the dead-end filtration properties based on the estimations of the cake porosity and the cake compressibility (Chun *et al*., 2004).

Accumulation of foulants is inevitable in inside-out dead-end filtration with hollow fibers and may be strongly affected by the flux level at which the filtration is carried out. There has been some indication in literature that by a smart choice of operational settings, irreversible fouling can be avoided. The critical filtered volume was found to depend on the applied flux. For the filtration of natural water, the reversibility of the formed cake depends on the filtrated volume as well (van de Ven *et al.,* 2008). Figure 2.4 shows the main flow direction for both dead end and cross-flow filtration.

In constant flux dead-end operation, the feed water is filtered for a given amount of time which is equivalent to a constant volume of permeate and a constant amount of fouling load assuming constant feed water composition. Due to the rejection of the matter dispersed and dissolved in the feed water, the resistance towards filtration increases. This in turn is generally related to concentration polarization, filter cake deposition and membrane fouling. To maintain a constant flux, the pressure has to be increased proportionally. All industrial plants operate at constant flux with an increasing feed pressure as a means to compensate for the increasing filtration resistance (van de Ven *et al*., 2008).

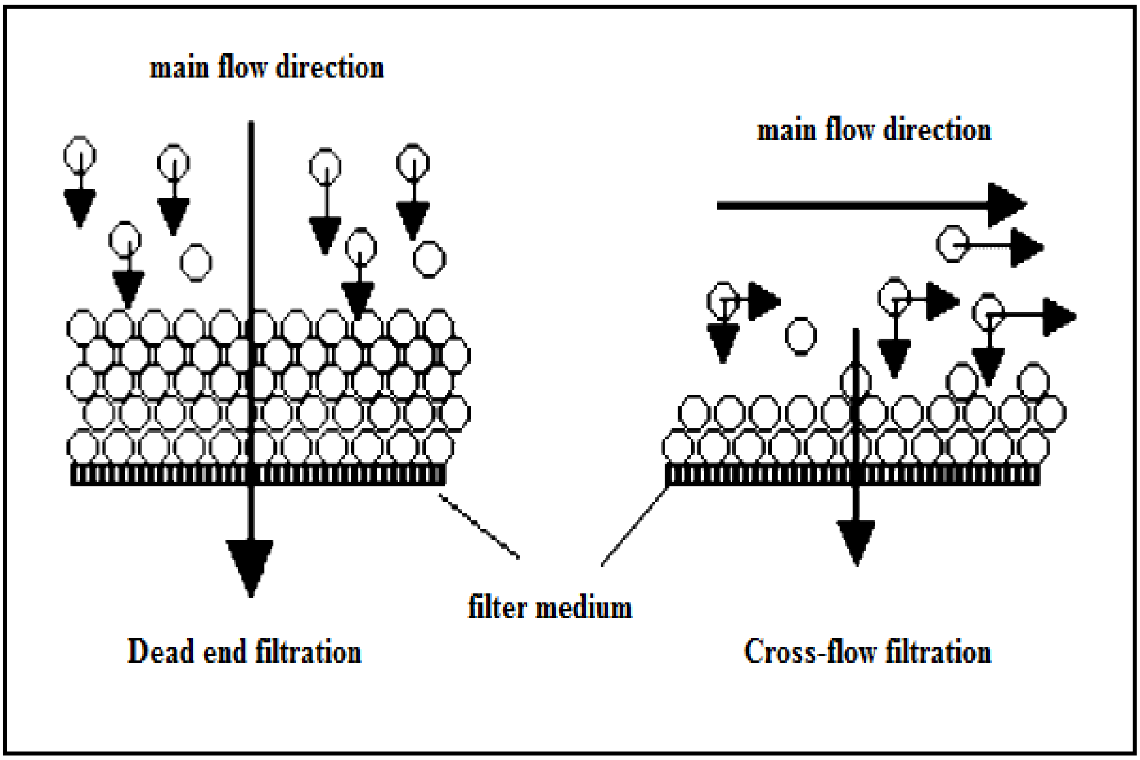


**Figure 2.4** Sketch of dead end and cross-flow filtration.

The tests carried out, have resulted in a preference for dead-end filtration over cross-flow mode. Not only does dead-end filtration have a lower energy consumption than cross-flow filtration, tests also turned out to result in a more stable process operation fluxes of 100 L/m h, at rather constant trans membrane pressure of 0.2– 0.6 bar (TMP), defined as head-loss across the membrane modules (Willemse and Brekvoort, 1999).

### 2.4 Membrane System

Membrane separation processes also widely used in various areas of chemical and fermentation processes. So, the membrane technology has been developed enormously. These is expressed in the vast amount of research which has gone into developing the right membrane type and module for different krids of separation process, developing new process as well as researches the best possible circumstances for the separation. These efforts have resulted in the present day commercialization of ultrafiltration (UF), microfiltration (MF), reverse osmosis or hyperfiltration (RO, gas separation, dialysis and electrodialysis (ED) (Derradji *et al*., 2005). This is because this process basically involves no phase change and chemical agents and is more environmentally friendly and economic (Juang *et al*., 2008).

Nowadays, membrane technologies are becoming more frequently used for separation of wide varying mixtures in the petrochemical-related industries and can compete successfully with traditional schemes (Ravanchi *et al*., 2009):

a) The technology behind membrane gas separation is potentially an energy-saving one, because the separation process takes place without phase transition.
b) It is also better for the environment, since the membrane approach requires the use of relatively simple and non-harmful materials.

c) The recovery of minor but valuable components from a main stream using membranes can be done without substantial additional energy costs.

d) Compared with conventional techniques, membranes can offer a simple, easy-to-operate, low-maintenance process option.

e) The development of novel materials for gas membrane manufacturing such as organic polymeric, hybrid organic-inorganic and inorganic will expand the use of membrane technology into new fields of applications in the petrochemical industry.

f) The process is simple.

g) There are diverse applications which can be studied by the same basic formulations.

h) The process is generally carried out at atmospheric conditions which, besides being energy efficient, can be important for sensitive applications encountered in pharmaceutical and food industry.

i) Modules can be added and optimized in a process design to achieve the desired separation.

j) Their systems have a low capital cost, compact size, modular configuration, and low specific power consumption, which reduce the production cost.

k) It is a clean process and requires simple and inexpensive filtration.

l) The process is continuous and the membranes do not require regeneration, unlike the adsorption or the absorption processes, which require a regeneration step leading to the use of two solid beds or a solvent regeneration unit.

In the wastewater treatment, membrane bioreactor technology has gained unprecedented popularity. Sludge, offers many advantages over conventional biological treatment such as small footprint, excellent treated water quality, and completes solids and liquid separation. One of the major obstacles for its widespread application is membrane fouling, which could cause severe loss of membrane permeability and thus the increase of energy consumption. The fouling phenomenon adversely affects both the quantity and quality of permeate flux (Lee *et al.*, 2001).